tissues, which are assumed to secret some factors under the regulation of nerves causing changes in the tissue stiffness. The tissues contain a large amount of the extracellular matrix mainly consisting of collagen fibrils, proteoglycans and microfibrils. The unique properties of these collagenous tissues might be due to lack of permanent associations between the collagen fibrils and the surrounding extracellular matrix. It seems that cross-linking between the fibrils are formed or broken during the change of the stiffness of the tissues. Its molecular mechanisms are, however, not yet fully understood. We isolated a protein factor called 'tensilin' from an extract of sea cucumber body wall dermis, one of the known catch connective tissues. It stiffens the detergent-treated dermal pieces and induces aggregation of collagen fibrils isolated from the tissue. We also isolated another protein factor which stiffens the dermal pieces. It is possible that there are other factors affecting on interactions among dermal fibrils and the stiffness of the tissues. Molecular mechanisms of the stiffness changes of the catch connective tissues should be clarified by purifying and characterizing these factors.

3711-Pos Board B439

Diffusion Discrepancy between Stroma of Tumor and Normal Tissues

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It is known that stromal microenvironments change in terms of its structure and composition during tumor progresses. Such change can lead to changes of diffusion efficiency and/or orientation of small molecules. Given small molecules such as cytokines and microRNAs are actively involved in tumorigensis, study diffusion in tumor stroma can lead to identifying the mechanism contributing to tumor progression. We used fluorescence recovery after photobleaching (FRAP) to examine the immobile fraction, diffusion rates, diffusion directionalities of dextran between 10 kD and 100 kD of molecular weight in stroma from both normal and tumor tissues from human breasts. We found that in the area with dense fibers, the diffusion rate in the tumor tissue is at least 2 fold-higher compared to the normal tissue. Furthermore, it was observed that 20% more dextran is immobilized in the tumor tissue, compared to the normal tissue, during the time frame of FRAP experiment, indicating the existence of efficient physical traps of small molecules in tumor stroma.

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Effect of Oligosaccharide Modified Material X on Viability of Human Cancer Cell Lines

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Anticarcinoma agents take part in the selective destruction of cancer cell lines, or inhibit the growth and proliferation of cancer cells. Finding anticarcinoma agents that have do not have noteworthy negative side effects is important matter for application in various flelds. Most of polysaccharides were used as medical product or an additive to health functional food. For example chitin and chitosan are known to exhibit antitumor, antibacterial, and antihypertensive activity. In this study, we incubated two kinds of cancer cells (Hep3B, A549) and 293T HEK cell treated with material X in concentration of 0.5%, 1% and 2% respectively for 24h. Then we measured viability of the cells by MTT assay. Our data suggest viability of 293T decreases gradually as concentration of X increases. Survival rates of 293T with X in concentration of 0.5%, 1% and 2% are 73.3%, 53.2% and 31.3% respectively. The cancer cell lines had more tolerance for 0.5% X and 1% X. However the cancer cells exhibited a rapid decline of viability when treated with 2% X. Survival rate of Hep3B with 2% X is 8.7% and that of A549 is 8.4%. With 2% X, cancer cell lines are about 4 times cytotoxic effects of the normal cell line. These results indicate that material X of specific concentration or higher depletes cancer cell lines while showing gradual effect on normal cell lines. To our knowledge, material X could be a promising antitumor application.

3713-Pos Board B441

Cytotoxic Effects of Substance a Obtained from Oligosaccharides on Human Lung Cancer Cell Line, A549

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It is reported that oligosaccharides have antitumoric effects on cancer cells. To evaluate the in vivo antitumor potentials of oligosaccharides, we obtained substance A from them. In this study, we treated three kinds of cell lines(Hep3B, A549, 293T) with substance A in concentration of 2%, 1%, 0.5% respectively and incubated them 24 hours. Then we used MTT assay to measure viability of cells. As a result, the viability of A549 cells decreased as the concentration of substance A became higher while other cells(Hep3B, 293T) were almost same. Survival rates of A549 cells were 84%, 82% and 68% when treated with substance A in concentration of 0.5%, 1% and 2% respectively. In all concentrations, cytotoxic effects of substance A on A549 cells were about 5 times stronger than on 293T cells. Our results suggested that substance A has selective cytotoxic effects on A549 cell lines. This study demonstrated that substance A has antitumor effects on human lung cancer.

3714-Pos Board B442

In Vivo Studies of Active Processes in the Escherichia Coli Nucleoid

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The cell is the site of actively motor-driven processes which drive the intracellular environment far from thermodynamic equilibrium. The dynamics of biological macromolecules such as DNA in such an environment are complex and subject to a multitude of constraints and forces. Inspired by our in vitro studies of DNA looping with optical tweezers that showed that additional non-thermal fluctuations in the DNA can substantially enhance the formation of regulatory DNA-protein complexes, we study the conformational fluctuations of chromosomal DNA in vivo in Escherichia coli by Fluorescence Correlation Spectroscopy (FCS).

Conformational fluctuations of the DNA move the bound fluorophores stochastically into the diffraction-limited excitation volume of a focused laser beam in a confocal microscope. From the time correlation functions of the measured fluorescence intensity, we quantify the fluctuations of the DNA as measured by its time-dependent mean square displacement, and the viscoelastic moduli of the nucleoid. These quantities in live cells significantly differ from the ATP-depleted dead cells on longer time scales, indicating that the fluctuations on longer time scale may be driven by active processes involving molecular motors that generate forces by ATP hydrolysis. On shorter time scales, we see little difference between live and dead cells, suggesting that the processes on corresponding short length scales rely primarily on thermally-driven diffusive mechanisms. We also note that the rheological properties of E. coli nucleoid significantly change when the ATP hydrolysis in cells is inhibited.

3715-Pos Board B443

The Energetic Contribution of Water in the Binding of Ribonuclease a and UMP

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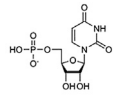
In most treatments of aqueous binding reactions, the energetic contribution of water is not addressed explicitly by the governing equation. The classical equation for a binding equilibrium ($\Delta G^\circ = -RT\ln K$) may be appropriate at infinite dilution but not under experimental conditions, especially in "nonideal" solutions containing other solutes. Resolving this issue is paramount to understanding the thermodynamics of molecular interactions in the context of a living cell. In the current study, we test a new equation that treats water as a co-reactant and co-product of the balanced reaction. The binding affinity of ribonuclease A (RNase A) with an inhibitor molecule, uridine-3'-monophosphate (UMP), is quantified using isothermal titration calorimetry. The results indicate that the equilibrium "constant," K, is dependent on reactant concentration and that the desolvation energy of binding is unfavorable for this specific protein-ligand interaction. These observations are consistent with published findings for another model binding system, the chelation of calcium by EDTA (*J. Phys. Chem. B* 2013, 117, 8180).


(RNase A)



(UMP)

3716-Pos Board B444

Size, Stoichiometry, and Organization of Soluble LC3-Associated Complexes

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